

## SEQUENCE LISTING

- <110> Quinnan, Gerald V. Zhang, Peng Fei
- <120> Expression and Characterization of HIV-1 Envelope Protein Associated with a Broadly Reactive Neutralizing Antibody Response
- <130> 44508-5001-US
- <140> US 09/762,261
- <141> 2001-02-05
- <150> US 60/095,267
- <151> 1998-08-04
- <150> PCT/US99/17596
- <151> 1999-08-04
- <160> 23
- <170> PatentIn Ver. 2.1
- <210> 1
- <211> 866
- <212> PRT
- <213> Human immunodeficiency virus type 1
- <220>
- <223> R2 strain envelope protein (gp 160)
- <400> 1
- Met Arg Val Lys Gly Ile Arg Arg Asn Tyr Gln His Trp Trp Gly Trp 15 10
- Gly Thr Met Leu Leu Gly Leu Leu Met Ile Cys Ser Ala Thr Glu Lys 30 20
- Leu Trp Val Thr Val Tyr Tyr Gly Val Pro Val Trp Lys Glu Ala Thr 45 35
- Thr Thr Leu Phe Cys Ala Ser Asp Ala Lys Ala Tyr Asp Thr Glu Ala 60 55 50
- His Asn Val Trp Ala Thr His Ala Cys Val Pro Thr Asp Pro Asn Pro 80 . 75 70 65

			٠.		•										
Gln_	Glu	Val	Glu	Leu	Val	Asn	Val	Thr	Glu	Asn	Phe	Asn	Met	Trp	Lys
 GIII_	GIU	Val	0.10	85					90					95	
Asn	λan	Met	Val	Glu	Gln	Met	His	Glu	Asp	Ile	Ile	Ser	Leu	Trp	Asp
ASII	ASII	Mec	100	014				105	-				110		
Cln	Car	I.e.ii	Taye	Pro	Cvs	Val	Lvs	Leu	Thr	Pro	Leu	Cys	Val	Thr	Leu
GIII	261	115	Буб	110	0,72		120					125			
7.00	Cvc	Thr	Λen	Len	Ara	Asn	Thr	Thr	Asn	Thr	Asn	Asn	Ser	Thr	Asp
ASII	130	1111	Ash	пси	9	135					140				
	<b>3</b>	7	Cor	Λαn	Car	Glu	Glv	Thr	Tle	Lvs	Gly	Gly	Glu	Met	Lys
Asn 145	Asn	ASI	Ser	ASII	150	Giu	Cly	1111	110	155	1	- 1			160
		_	<b>m</b> 1	3	T1.	Ala	Thr	Sar	Tle	Glv	Asp	Lvs	Met	Gln	Lys
Asn	Cys	Ser	Pne	165	116	Ala	1111	361	170			-1 -		175	-
		_ •	_	•	<b></b>	Trea	T 011	Λαn	Tle	Glu	Pro	Tle	. Asp	Asn	Asp
Glu	Tyr	Ala	_Leu 180		Tyr	пув	пеи	185		- OI u	110		190		Asp
					_	<b>~</b> 1 -	0	Crra	Nan	Thr	· Ser	· Val	Tle	Thr	Gln
Asn	Thr	Ser 195		Arg	ьeu	ile	200	Cys	ASII	1 1111	DCI	205	5		Gln
									<b>T</b> ]_	Dage	. Tla	. uic	• ጥኒ/ን	- Cvs	Δla
Ala	Cys 210		Lys	Ile	Ser	Phe 215		Pro	116	PIC	220	) )	, 1y1	. Cyr	Ala
					_ •	_	_		. 3	. 7. ~ ~	. T.,	1 T.176	- Dhe	Ser	- Glv
Pro 225		Gly	√ Ph∈	e Ala	1le 230		Lys	Cys	ASI	235	<u>.</u> Э пЪя	з пув	5 F110	5 561	Gly 240
												. mb.	~ TT-1 0	. (1)	, Tla
Lys	Gly	seı Seı	Cys	Lys 245		ı Val	Ser	Thi	250°	C Gir	ı Cys	i Tni	r His	255	/ Ile
												_	_	<b>7</b> 7.	- 01
Arg	Pro	Va:	L Va 260		Thr	: Gln	Let	Let 26!	ı Leı 5	ı Ası	n Gly	y Sei	r Lei 27	) J Ale	a Glu
												÷			<b>—</b> 1
Glu	Glu			l Ile	e Arg	g Ser	Gli 280		n Phe	e Thi	r Ası	n Ası 28	n Ala 5	а Бу	s Thr
		27													
Ile			l Gl	n Lei	ı Arg	g Glu 295		va:	l Ly:	s Il	e Asi	n Cy 0	s Se	r Ar	g Pro
	290													_	_
		n As	n Th	r Ar			c Ile	e Pr	o Me	t Gl; 31	y Pr	o Gl	y Ar	g Al	a Phe 320
305					31										
Туз	Th	r Th	r Gl			e Ile	e Gl	y As			g Gl	n Al	a Hi	s Су 33	s Asn 5
				32	5				33	U				,,	_

Ile_	Ser	Lys	Thr	Asn	Trp	Thr	Asn	Ala	Leu	Lys	Gln	Val	Val	Glu	Lys
			340					345					350		
Leu	Gly	Glu 355	Gln	Phe	Asn	Lys	Thr 360	Lys	Ile	Val	Phe	Thr 365	Asn	Ser	Ser
Gly	Gly 370	Asp	Pro	Glu	Ile	Val 375	Thr	His	Ser	Phe	Asn 380	Cys	Ala	Gly	Glu
Phe 385	Phe	Tyr	Cys	Asn	Thr 390	Thr	Gln	Leu	Phe	Asp 395	Ser	Ile	Trp	Asn	Ser 400
Glu	Asn	Gly	Thr	Trp 405	Asn	Ile	Thr	Arg	Gly 410	Leu	Asn	Asn	Thr	Gly 415	Arg
Asn	Asp	Thr	Ile 420	Thr	Leu	Pro	Cys	Arg 425	Ile	Lys	Gln	Ile	Ile 430	Asn	Arg
Trp	Gln	Glu 435	Val	Gly	Lys	Ala	Met 440	Tyr	Ala	Pro	Pro	Ile 445	Lys	Gly	Asn
Ile	Ser 450		Ser	Ser	Asn	Ile 455	Thr	Gly	Leu	Leu	Leu 460	Thr	Arg	Asp	Gly
Gly 465		Asp	Asp	Asn	Ser 470	Arg	Asp	Gly	Asn	Glu 475	Thr	Phe	Arg	Pro	Gly 480
Gly	Gly	Asp	Met	Arg 485	Asp	Asn	Trp	Arg	Ser 490		Leu	Tyr	Lys	Tyr 495	Lys
Val	Val	. Lys	500		Pro	Leu	Gly	Val		Pro	Thr	Lys	Ala 510	Lys	Arg
Arg	<sub>[</sub> Val	. Val		n Arg	g Glu	Glu	Arg		Val	Gly	Leu	Gly 525		Met	Phe
Ph∈	61 530		e Lev	ı Gly	⁄ Ala	Ala 535		Ser	Thr	Met	Gly 540		Ala	Ser	Val
Thr 545		ı Thi	. Val	l Glr	n Ala 550		g Glr	ı Leu	Leu	Ser 555		Ile	· Val	Gln	Gln 560
Glr	n Sei	r Ası	ı Le	ı Lei 565		j Ala	a Ile	e Glu	1 Ala 570		Gln	His	. Leu	Lev 575	Gln
Let	ı Th:	r Vai	l Trj 58		/ Ile	e Lys	∈ Glr	1 Leu 585		n Ala	a Arg	, Ile	590	ı Ala	va'l

Glu Arg Tyr Leu Lys Asp Gln Gln Leu Leu Gly Ile Trp Gly Cys Ser Gly Lys Leu Ile Cys Thr Thr Thr Val Pro Trp Asn Ala Ser Trp Ser Lys Asn Lys Thr Leu Glu Ala Ile Trp Asn Asn Met Thr Trp Met Gln Trp Asp Lys Glu Ile Asp Asn Tyr Thr Ser Leu Ile Tyr Ser Leu Ile Glu Glu Ser Pro Ile Gln Glu Lys Asn Glu Gln Glu Leu Leu Glu Leu Asp Lys Trp Ala Asn Leu Trp Asn Trp Phe Asp Ile Ser Asn Trp Leu Trp Tyr Ile Lys Ile Phe Ile Met Ile Val Gly Gly Leu Val Gly Leu Arg Ile Val Phe Val Val Leu Ser Ile Val Asn Arg Val Arg Gln Gly Tyr Ser Pro Leu Ser Phe Gln Thr Arg Leu Pro Ala Pro Arg Gly Pro Asp Arg Pro Glu Glu Ile Glu Glu Glu Gly Gly Asp Arg Asp Arg Asp Arg Ser Gly Leu Leu Val Asp Gly Phe Leu Thr Leu Ile Trp Val Asp Leu Arg Ser Leu Cys Leu Phe Ser Tyr His Arg Leu Arg Asp Leu Leu Leu Ile Val Thr Arg Ile Val Glu Leu Leu Gly Arg Arg Gly Trp Glu Ile Leu Lys Tyr Trp Trp Asn Leu Leu Gln Tyr Trp Ser Gln Glu Leu Lys Asn Ser Ala Val Ser Leu Phe Asn Ala Thr Ala Ile Ala Val Ala Glu Gly Thr Asp Arg Val Ile Gln Val Leu Gln Arg Val Gly Arg

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Ala Leu Leu His Ile Pro Thr Arg Ile Arg Gln Gly Leu Glu Arg Ala
                                              860
                          855
  Leu Leu
  865
  <210> 2
  <211> 17
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  <213> Human immunodeficiency virus type 1
  <220>
  <223> segment of R2 strain V3 domain
  <400> 2
  Lys Ser Ile Pro Met Gly Pro Gly Arg Ala Phe Tyr Thr Thr Gly Gln
                                        10
  Ile
   <210> 3
   <211> 35
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   <223> R2 strain V3 domain
   <400> 3
   Cys Ser Arg Pro Asn Asn Asn Thr Arg Lys Ser Ile Pro Met Gly Pro
                                        10
     1
Gly Arg Ala Phe Tyr Thr Gly Gln Ile Ile Gly Asp Ile Arg Gln
                                                         30
                                    25
                20
   Ala His Cys
   <210> 4
   <211> 35
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   <220>
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<223> V3 domain of strain 93TH966.8
<400> 4
Cys .Thr Arg Pro Ser Asn Asn Thr Arg Thr Ser Thr Thr Ile Gly Pro
                                     10
                  5
Gly Gln Val Phe Tyr Arg Thr Gly Asp Ile Thr Gly Asn Ile Arg Lys
                                                      30
             20
Ala Tyr Cys
         35
<210> 5
<211> 13
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: derivatives
      of segment of V3 domain in R2 strain
<220>
<221> VARIANT
<222> (3)..(12)
<223> X = any natural or non-natural amino acid.
 <400> 5
Pro Met Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Gln
                   5
 <210> 6
 <211> 30
 <212> PRT
 <213> Human immunodeficiency virus type 1
 <220>
 <223> sequence of Phenetic 1 in V3 region
 <400> 6
 Asn Asn Thr Arg Lys Ser Ile His Ile Gly Pro Gly Arg Ala Phe Tyr
 Thr Thr Gly Asp Ile Ile Gly Asp Ile Arg Gln Ala His Cys
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<210> 7
<211> 30
<212> PRT
<213> Human immunodeficiency virus type 1
<220>
<223> sequence of Phenetic 2 in V3 region
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Asn Asn Thr Arg Lys Ser Ile Ser Ile Gly Pro Gly Arg Ala Phe Tyr
                                      10
                  5
Ala Thr Gly Glu Ile Ile Gly Asp Ile Arg Gln Ala His Cys
                                  25
             20
<210> 8
<211> 30
<212> PRT
<213> Human immunodeficiency virus type 1
 <220>
 <223> sequence of Phenetic 3 in V3 region
 <400> 8
 Asn Asn Thr Arg Lys Ser Ile Ser Ile Gly Pro Gly Arg Ala Phe Tyr
                                                           15
                   5
 Ala Thr Gly Lys Ile Ile Gly Asp Ile Arg Gln Ala His Cys
 <210> 9
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 <212> PRT
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 <223> sequence of Phenetic 4 in V3 region
  <400> 9
 Asn Asn Thr Arg Lys Ser Ile Arg Ile Gly Pro Gly Gln Ala Phe Tyr
                    5
  Ala Thr Gly Asp Ile Ile Gly Asp Ile Arg Gln Ala His Cys
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<210> 10
<211> 30
<212> PRT
<213> Human immunodeficiency virus type 1
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<223> sequence of Phenetic 5 in V3 region
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Asn Asn Thr Arg Lys Ser Ile His Ile Gly Pro Gly Arg Ala Phe Tyr
                                      10
                  5
Ala Thr Gly Lys Ile Ile Gly Asp Ile Arg Gln Ala His Cys
                                  25
              20
 <210> 11
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 <400> 11
 Lys Asn Thr Arg Arg Arg Ser His Ile Gly Pro Gly Arg Ala Phe Tyr
                    5
 Thr Thr Lys Gln Ile Ile Gly Asp Ile Arg Gln Ala His Cys
                                   25
  <210> 12
  <211> 30
  <212> PRT
  <213> Human immunodeficiency virus
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  <223> sequence of Phenetic 7 in V3 region
  Asn Asn Thr Arg Thr Ser Ile Thr Ile Gly Pro Gly Gln Val Phe Tyr
  <400> 12
                                        10
  Arg Thr Gly Lys Ile Ile Gly Asp Ile Arg Gln Ala His Cys
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<210> 13
<211> 30
<212> PRT
<213> Human immunodeficiency virus type 1
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<223> sequence of Phenetic 8 in V3 region
Lys Lys Met Arg Thr Ser Ala Arg Ile Gly Pro Gly Arg Val Phe His
 Lys Thr Gly Asp Ile Ile Gly Ser Ile Thr Lys Ala Tyr Cys
                                  25
 <210> 14
 <211> 29
  <212> PRT
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  <220>
  <223> sequence of Phenetic 9 in V3 region
  Asn Asn Thr Arg Gln Ser Thr His Ile Gly Pro Gly Gln Ala Leu Tyr
  Thr Thr Asp Ile Ile Gly Lys Ile Arg Gln Ala His Cys
                20
   <210> 15
   <211> 29
   <212> PRT
   <213> Human immunodeficiency virus type 1
   <220>
   <223> sequence of Phenetic 10 in V3 region
    Asn Asn Thr Arg Gln Gly Thr His Ile Gly Pro Gly Arg Ala Tyr Tyr
    Thr Thr Asn Ile Ile Gly Asp Ile Arg Gln Ala His Cys
```

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<210> 16
<211> 29
<212> PRT
<213> Human immunodeficiency virus
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Asn Asn Thr Arg Gln Arg Thr Ser Ile Gly Gln Gly Gln Ala Leu Tyr
 Thr Thr Glu Ile Arg Gly Asp Ile Arg Gln Ala Ala Cys
              20
  <210> 17
  <211> 30
  <212> PRT
  <213> Human immunodeficiency virus type 1
  <223> sequence of Phenetic 12 in V3 region
   Asp Asn Ile Lys Ile Gln Arg Thr Pro Ile Gly Gln Gly Gln Ala Leu
     1
   Tyr Thr Thr Arg Ile Thr Gly Tyr Ile Gly Gln Ala His Cys
                20
    <210> 18
    <211> 29
    <213> Human immunodeficiency virus type 1
    <212> PRT
    <223> sequence of Phenetic 13 in V3 region
     Gln Asn Lys Arg Gln Gly Thr Pro Ile Gly Leu Gly Gln Ala Leu Tyr
       1
     Thr Thr Arg Ile Lys Gly Asp Ile Arg Lys Ala His Cys
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<210> 19
<211> 30
<213> Human immunodeficiency virus type 1
<223> sequence of Clade A in V3 region
 Asn Asn Thr Arg Lys Ser Val His Ile Gly Pro Gly Gln Ala Phe Tyr
                   5
   1
 Ala Thr Gly Asp Ile Ile Gly Asp Ile Arg Gln Ala His Cys
  <210> 20
  <211> 30
  <213> Human immunodeficiency virus type 1
   <223> sequence of Clade B in V3 region
   Asn Asn Thr Arg Lys Ser Ile His Ile Gly Pro Gly Arg Ala Phe Tyr
                     5
      1
    Thr Thr Gly Glu Ile Ile Gly Asp Ile Arg Gln Ala His Cys
                 20
    <210> 21
     <211> 30 ....
     <213> Human immunodeficiency virus type 1
     <212> PRT
     <223> sequence of Clade C in V3 region
      Asn Asn Thr Arg Lys Ser Ile Arg Ile Gly Pro Gly Gln Thr Phe Tyr
                       5
        1
      Ala Thr Gly Asp Ile Ile Gly Asp Ile Arg Gln Ala His Cys
                    20
```

<210> 22 <211> 29 <212> PRT <213> Human immunodeficiency virus type 1 <220> <223> sequence of Clade D in V3 region Asn Asn Thr Arg Gln Arg Thr His Ile Gly Pro Gly Gln Ala Leu Tyr Thr Thr Arg Ile Ile Gly Asp Ile Arg Gln Ala His Cys 20 <210> 23 <211> 30 <212> PRT <213> Human immunodeficiency virus type 1 <220> <223> sequence of Clade E in V3 region Asn Asn Thr Arg Thr Ser Ile Thr Ile Gly Pro Gly Gln Val Phe Tyr 1 Arg Thr Gly Asp Ile Ile Gly Asp Ile Arg Lys Ala Tyr Cys 20